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REVIEW ARTICLE

Current Understanding in the Pathophysiology of SARS-CoV-2-Associated Rhino-Orbito-Cerebral Mucormycosis: A Comprehensive Review

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Abstract
Aim Recently, with the second wave of COVID-19, the Indian subcontinent has witnessed a dramatic rise in mucormycosis infection in patients recovered from COVID-19. This association has been documented in various case reports/case series and institutional experiences, and the mortality associated with this fungal infection is emerging as a cause of concern. The aim of the present paper is to provide a scientific overview on the pathogenesis of mucormycosis in COVID-19 beyond the conventional understanding of the disease process, which may not otherwise explain the increased incidence of mucormycosis in SARS-CoV-2.
Methodology This paper is structured as a narrative review of the published literature on the pathogenesis of COVID-19 which contributes to the development of mucormycosis. Apart from the acknowledged role of ketoacidosis, high blood sugar, and iron metabolism in the pathogenesis of mucormycosis, other factors involved in pathophysiology of COVID-19 which might alter or enhance the mucormycosis infection such as (1) the role of ferritin, (2) high serum iron, (3) free radical-induced endothelitis, (4) hepcidin activation, (5) upregulation of glucose receptor protein (GRP78) are discussed in the pathophysiology of COVID-19-associated mucormycosis.
Conclusion A new proposal for the pathogenesis based on the ferritin, viral mimicry of hepcidin and GRP78–CotH3 interaction, which clearly explains the surge in mucormycosis in SARS-CoV-2 infection, has been explained.

Keywords Mucormycosis · COVID-19 · Ferritin

Introduction

Mucormycosis is a highly invasive fungal infection afflicting predominantly the immunocompromised patients. The genera responsible for human infection are Rhizopus, Mucor and Rhizomucor; Cunninghamella, Lichtheimia and Apophysomyces [1]. This fungi are ubiquitous in nature, and human infections are rare. The principal risk factors for mucormycosis include diabetes mellitus (DM), with or without ketoacidosis, hematological malignancies, organ transplant, chronic kidney or liver disease, immunological disorders, prolonged corticosteroid therapy, deferoxamine therapy and trauma [1, 2].

The incidence of mucormycosis has seen an upsurge in the last decade in both developed and developing countries and is still rising; however, the rise in Asia and India specifically has been alarming. In India, the prevalence of mucormycosis is approximately 0.14 cases per 1000 population, which is about 80 times the prevalence of mucormycosis in developed countries [3]. Factors which contribute to the pathogenesis and virulence nature of Mucor include rapid growth, ability to utilize the host iron for growth, ability to adhere to the endothelial surface and downregulation of host defense genes responsible for immune defense [4, 5]. Based on the clinical involvement,

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it is classified into rhino-cerebral, pulmonary, cutaneous, disseminated and gastrointestinal [5].

The prognosis of mucormycosis is relatively poor especially when the host is immunocompromised and when the diagnosis is delayed. The overall survival rates of rhino-cerebral disease [50%] are far better than its pulmonary and disseminated counterparts partly because of its early diagnosis [6]. The general consensus in the management of rhino-cerebral mucormycosis is surgical debridement followed by antifungal therapy [6].

COVID-19 is an infective inflammatory disease with multisystem involvement caused by SARS-CoV-2. The major factors involved in the pathophysiology of SARS-CoV-2 disease are (i) markedly raised proinflammatory CD4 T cells and CD8 toxic granules, (ii) cytokine surge, (iii) hemoglobinopathy, (iv) hypercoagulable state, (v) altered iron metabolism and increased iron overload resulting in hypoxia and multisystem failure in severe cases [5, 7, 8].

Recently, with the second wave of COVID-19, the Indian subcontinent has witnessed a dramatic rise in mucormycosis infection in patients recovered from COVID-19. This association has been documented in various case reports/case series and institutional experiences [6, 9, 10]. Furthermore, mortality associated with this fungal infection is emerging as a cause of concern in Indian subcontinent. Therefore, the aim of the present paper is to provide a scientific overview on the pathogenesis of mucormycosis in COVID-19 beyond the conventional understanding of the disease process, which may not otherwise explain the increased incidence of fungal infection predominantly the mucormycosis in SARS-CoV-2.

Apart from the acknowledged role of ketoacidosis, high blood sugar and iron metabolism in the pathogenesis of mucormycosis, other factors involved in pathophysiology of COVID-19 which might alter or enhance the mucormycosis infection like (i) the role of ferritin, [ii] high serum iron, (iii) free radical-induced endothelitis, (iv) hepcidin activation by viral mimicry, (v) upregulation of glucose receptor protein (GRP78) are discussed in the pathophysiology.

Is Viral-Induced Immune System Dysfunction the Cause for Mucormycosis?

The inoculation of SARS-CoV-2 into the host results in the activation of innate immune system. However, the mechanism by which immune system responds to the virus is just unfolding, which limits our current understanding of possible immune-mediated pathways facilitating the pathogenesis of mucormycosis. Clinical evidence suggests that the neutrophils monocytes and macrophages which plays a predominant role in the primary host defense against Mucorales are unaffected in COVID-19 infection, thus eliminating their role in the pathogenesis [11].

On the contrary, an increase in peripheral neutrophil number was noted in COVID-19 with an increased neutrophil lymphocyte ratio [12]. This is in fact beneficial as far as immunity toward Mucorales is concerned. These neutrophils are very effective and readily inactivate the fungus by the generation of oxidative metabolites if the host is immunocompetent.

Analyzing the existing literature lymphopenia seems to be the only significant immune cell defect detected in COVID-19 [11]. However, lymphopenia does not play any significant role in increasing the host susceptibility to Mucorales. Clinically, this can be explained by the lower incidence of mucormycosis in HIV-infected patients and other lymphopenic syndromes. A retrospective study has shown only 2 cases of mucormycosis in autopsy of 1630 patients died of AIDS-related complications from 1984 to 2002, signifying the rarity of incidence [13].

Mucorales are ubiquitous fungi with various modes of entry into the host tissues. The nasal mucosa, sinus, skin and endothelium constitute the fundamental barriers against fungal invasion. Any injury, chronic irritation or breach in continuity (nasal prongs, swab stick injury) of nasal or sinus mucosa would result in the adherence of fungus to the laminin and type IV collagen on the basal cell layer of epithelium [14].

Keeping this in view, on reviewing the existing literature on COVID-19, it has been observed that nasal mucociliary clearance was profoundly delayed throughout the course of infection and may persist even after [15, 16]. The nasal mucociliary clearance is the primary innate defense mechanism of the paranasal sinus against various pathogens. This mechanism protects the upper respiratory system from various inhaled particles and microorganisms. However, the significance of delayed mucociliary clearance independently as a potential cause for the development of Mucor is debatable. Nevertheless, it becomes one of the critical factors for the growth of fungus together with the inflammation of the upper airway, in patients requiring prolonged hospital stay with supplemental oxygen therapy.

Since thrombosis is a hallmark of mucormycosis, platelets do play an important role in the innate immunity against the fungus. Mild thrombocytopenia is detected in 58–95% of patients with SARS-CoV-2 infection [17]. A few suggested mechanisms through which platelets possibly hamper the growth of fungi are (i) directly by adhering to Mucorales hyphae to form thrombus. (ii) Indirectly, platelets secrete a battery of proinflammatory and anti-inflammatory cytokines such as TGF-β and Thrombocidins which may act against Mucorales. Additionally, CD154 and platelet Toll-like receptors are present on platelet membrane which facilitate platelet binding to cells such as

endothelial cells, dendritic cells, monocytes and T and B lymphocytes leading to their activation [18]. However, to consider mild thrombocytopenia as potential clinical predictor of the disease may not be right and lacks congruity.

Studies have shown that the incidence of candidiasis and aspergillosis is 50-fold more common than mucormycosis [19]. Candida is a well-known commensal in the human body with marked capabilities to go virulent. This virulent attribute enables Candida to emerge as the most recovered pathogen in intensive care units and undoubtedly the most common infective fungal disease with incidence ranging from 6 to 10% in all critical care patients [19]. In the present scenario, a remarkable proportion of COVID-19 patients develop respiratory complications and a majority of them often require long-term mechanical ventilation/antibiotics/steroids as a part of the management protocol. This predisposes the host to various nosocomial infections/supra-infections [bacterial or fungal] particularly if the host is immunocompromised.

Essentially the hazards of long-term antibiotics, steroid and mechanical ventilation on the host are undisputable. While candidiasis and aspergillosis are the most common opportunistic infections affecting such patients, mucormycosis has largely been a rare occurrence. This clearly indicates the presence of alternative factors apart from the overt immune dysfunction as a chief cause for development of mucormycosis in SARS-CoV-2.

Diabetes Mellitus, Virus-Mediated Pancreatic Damage and Corticosteroid Usage

Fungal inoculation into the host tissues results in the activation of both innate and adaptive immune responses. However, in the presence of hyperglycemia the innate immune system is defective resulting in the inhibition of neutrophil migration, chemotaxis and decreased phagocytosis. Diabetes with ketoacidosis [DKA] is 50 percent more likely to develop mucormycosis than without DKA [2]. Ketone bodies [beta-hydroxybutyrate] by virtue of its high pH increase the availability of free iron by inhibiting the sequestration of iron by transferrin and ferritin. This high pH and increased availability of free iron promote fungal growth in a susceptible host. Similarly at a physiological concentration of ketone bodies, expression of GRP78 [Glucose Regulator Protein 78] in endothelium and fungal protein CotH3 is increased [20].

In the absence of DKA, hyperglycemia also increases the risk of mucormycosis by the following mechanisms (i) inhibiting the action of iron sequestrating proteins, (ii) upregulation of GRP78 and fungal protein, CotH3, (iii) impaired phagocytosis and chemotaxis by neutrophils and (iv) by weakening the oxidative and nonoxidative pathways [2]. While examining the existing literature, one would observe a definite correlation between the underlying comorbid condition and the type of mucormycosis. Rhino-cerebral mucormycosis is almost always associated with diabetic ketoacidosis. While hematological malignancies and neutropenia cause pulmonary disease, trauma usually leads to cutaneous mucormycosis [2].

It is confirmed that the patients with ketoacidosis are at a higher risk of developing rhino-cerebral mucormycosis [8]. The mechanisms which predispose the diabetic ketoacidosis to rhino-cerebral disease are obscure. Perhaps the acidic pH of DKA and high free iron have a role to play. Mucorales exclusively depend on the host iron for their metabolic requirements. However, many authors find this inference not convincing mainly because of the chief presentation of mucormycosis in patients taking iron chelating agents like deferoxamine, wherein the fungus uses the chelated iron as a siderophore for its growth is exclusively, disseminated type [21]. This establishes the fact that high serum iron cannot by itself explain the predisposition of rhino-cerebral disease with DKA. Similarly, other observations in DKA like impaired neutrophil chemotaxis and phagocytosis cannot be consistently applied to the increased rhino-cerebral cases due to the fact that neutropenic patients frequently develop pulmonary and disseminated disease rather than rhino-cerebral disease [8].

Although the mechanism is not fully understood, evidence suggests that SARS-CoV-2 infection impairs the function of pancreatic beta cells resulting in acute DKA. It is interesting to note that the ketoacidosis in euglycemic states was also noted in many patients throughout the infection [22]. Smith et al. have observed that 29 euglycemic patients with normal HbA1C levels developed hyperglycemia during course of COVID-19 indicating a possible damage to the pancreatic cells [23].

In COVID-19 patient's studies have shown that the glycemic control is not only poor but also necessitates insulin to be utilized in exceedingly high doses for their management. This transient elevation of glucose during the course of disease can be possibly linked to insulin resistance caused by increased levels of inflammatory cytokines in the body [24, 25].

Although DKA is normally seen in T1DM diabetes mellitus, a recent systematic review on COVID-19 has shown that majority of diabetic ketoacidosis was observed with T2DM [77%] cases [26]. An analysis of similar studies has revealed that COVID-19 can precipitate two categories of T1DM in patients [27]. The first group manifests T1DM with ketoacidosis that occurs at the onset of COVID-19 infection, and the second group exhibits T1DM without ketoacidosis in the initial stages, but in whom ketoacidosis occurred several weeks after apparent recovery from COVID-19 [28]. In addition to that, on

reviewing the literature on the recovery of patients from SARS-CoV-1 in the past, it was observed that hyperglycemia persisted for approximately three years indicating a long-term damage to pancreatic β-cells [29].

Hence, it may be summarized that SARS-CoV-2 could possibly trigger diabetes with ketoacidosis at least in a small subset of population and may even persist several weeks or months after apparent recovery from the disease. This could possibly corroborate the late onset of mucormycosis in a few clinical series usually weeks after the recovery from COVID-19. Therefore, in line with the evidence, the hyperglycemia, ketoacidosis, increased availability of free iron and impaired phagocytic action invariably leads to an environment conducive for the growth of fungi. Similarly, increased serum iron and overexpression of GRP78 in DKA cause the endothelial damage predisposing the host to fungal invasion. Nonetheless, a larger epidemiological studies are required to validate the link between new onsets 'COVID-diabetes' as a contributing factor in the pathogenesis of mucormycosis in SARS-CoV-2.

Corticosteroids

Apart from the viral-induced hyperglycemia, systemic steroids and antiviral agents used in the management of COVID-19 can also be considered as contributing factors in the worsening of hyperglycemia. There is an increased incidence of mucormycosis infections in diabetic patients treated for COVID-19, who received corticosteroid administration during the course of treatment [9]. Corticosteroids and immunosuppressive agents are risk factors and prolonged high dose (> 3 weeks) of corticosteroids predisposes an individual to angioinvasive Mucormycosis infection [30]. The propensity of corticosteroids to impair migration, ingestion and phagolysosome fusion in macrophages may explain suppressed immunity in such patients. There is a positive correlation between coronavirus and mucormycosis of the paranasal sinuses (ethmoidal sinus in particular) which must be taken into consideration.

Min et al. evaluated the adverse effect of short-term high-dose steroid retrospectively in a cohort of 500 patients who were given 48 mg/day or higher (for Bell's palsy, sudden sensorineural hearing loss and Ramsay Hunt syndrome) for a period of two weeks [31]. The adverse effects were seen in one-third of the patients but was limited to abdominal discomfort, skin rash and hot flush. No cases of infection were noted throughout the study period. Undeniably, long-term corticosteroids, irrespective of the dose, predispose the patients to a variety opportunistic infections. However, as evident in the literature, the occurrence of mucormycosis prior to/without candidiasis or aspergillosis as the primary opportunistic pathogen in a host with relatively short duration of steroid therapy (< 2 weeks) is mutually contradictory.

The Viral Modulation of Iron Metabolism—role of Ferritin, Viral Mimicry of Hepcidin and 'Endothelitis'

The accusation of host iron is one of the most important virulent traits of Mucorales. In humans, serum iron is bound by iron storage proteins like transferrin and ferritin making free iron unavailable to pathogens [32]. This sequestration of iron constitutes an important host defense mechanism against Mucorales. Recent studies have demonstrated that the *R. oryzae* grows poorly in iron deficient medium elucidating the importance of iron in the growth and metabolism of Mucorales [33].

Dysregulation of iron homeostasis and iron overload is a notable factor in the pathogenesis of SARS-CoV-2 [8, 35]. During active infection, the viral particle interacts with hemoglobin molecule through the ACE2 and CD147 receptors and subsequently undergoes viral endocytosis through the spike protein on the viral surface SARS-CoV-2 [7]. This would perpetuate a cascade events resulting in dysfunctional hemoglobin, hemolysis, accumulation of heme and reduced oxygen transportation [34, 35].

During SARS-CoV-2 infection, an increased serum ferritin is released into the circulation in response to the inflammatory response [36, 37]. Ferritin is a clinical marker of acute inflammation and plays a vital role in the pathogenesis of COVID-19 [38]. The principal function of ferritin appears to be the storage of iron, thus protecting the cells from excessive free iron and its toxic effects. Soon after the release of ferritin, it loses a part of its inner iron content resulting in an elevated level of free serum iron [39]. Excessive free iron in the serum further signals the liver to increase the production of more ferritin [40].

Furthermore, macrophage activation and increased IL-6 secretion because of the disease progression contribute to 'hyperferritinemia' accentuating this vicious cycle [5, 36]. Additionally, the excessive intracellular free iron generates reactive oxygen species by Fenton reaction leading to oxidative stress and lipoperoxidation of cell membranes [41]. This free radical-mediated endothelial destruction catalyzed by the presence of free iron and ferritin results in a diffuse inflammation of endothelium leading to 'Endothelitis' [7, 41].

Worsening the existing condition, the hepcidin mimetic effects exhibited by the SARS-CoV-2 virus also increase the ferritin independent of the inflammatory response [7]. Hepcidin is the main regulator of iron metabolism as it interacts with ferroportin, another transport protein to

transport iron inside the cells [42]. Ehsani et al. have emphasized the similarity between SARS-CoV-2 spike glycoprotein and the hepcidin [43]. In a complex mechanism mimicking the hepcidin, SARS-CoV-2 uses its spike proteins to invade the cytoplasm. This hepcidin mimicking activity of SARS-CoV-2 causes dysregulation of iron metabolism leading to subsequent hyperferritinemia and ferroptosis [44]. Besides, the presence of systemic inflammation and elevated levels of cytokines assists in hepcidin upregulation and ferroportin downregulation causing a significant increase in intracellular free iron.

In vitro studies have shown that the growth curve of Rhizopus is directly proportional to the presence of free iron in the serum [33, 45]. Additionally, animal studies have demonstrated decreased survival rate and poor therapeutic outcome when free iron was added in the experimental animal. Moreover, the outcomes of the only randomized control study ever conducted for the management of mucormycosis suggested that higher baseline blood iron levels were predictive of higher risk of mortality. On that account, it is logical to consider iron chelation as an adjunctive therapy in the management of mucormycosis. This is particularly important as the virulence of *R. oryzae* depends on the ability of fungus to gather free iron from the host. Again, this unique feature differentiates Mucorales from other opportunistic fungi like Candida and Aspergillosis [2]. Iron chelators like deferasirox and deferiprone may prove to be beneficial in many clinical studies [21, 46, 47]. In addition to the binding with the free iron, the iron chelators also help in the downregulation of hepcidin, thus counteracting the hyperferritinemic syndromes [5].

Notwithstanding the acknowledged role of ketoacidosis, high blood sugar and iron metabolism in the pathogenesis of mucormycosis, other factors involved in SARS-CoV-2 pathophysiology which might modify or enhance the pathogenesis of Mucorales are high ferritin, increased serum iron, endothelitis, hepcidin activation and upregulation of GRP78 receptors. Arguably all these predisposing factors are primarily mediated by the SARS-CoV-2 infection implying a possible association between high incidence of mucormycosis and COVID-19 patients.

Fungal Interaction with Host Tissue: Angioinvasion: The Role of SARS-CoV-2-Mediated Endothelitis and Upregulation of GRP78 and CotH3

The distinctive pathognomic feature of mucormycosis is its potential to invade the blood vessels, causing vessel thrombosis and subsequent tissue necrosis [48]. Once the fungus enters the mucosa or skin, it adheres to the extracellular matrix protein laminin, present on the basement membrane [14]. Consequently, the Mucor interacts with the endothelial cells by the help of a specific receptor GRP78 present on the host endothelium facilitating fungal endocytosis. The fungal ligand which helps in binding the GRP78 during early invasion of Mucor belongs to a spore coating protein family (CotH3) [49].

GRP78 is a class of heat shock protein released from the endoplasmic reticulum of the endothelial cells in response to stress [50]. During SARS-CoV-2 infection, the viral spike glycoprotein triggers the endoplasmic reticulum stress [ERS] and stimulates the synthesis of GRP78 [51]. A prospective case control study reveals that the serum GRP78 was 5 times higher in COVID-19 as compared to the control group [52, 53]. Furthermore, studies have demonstrated that the SARS-CoV-2 virus also uses the same GRP78 for its internalization into the host cells [51]. By upregulating the GRP78 by the virus for its own entry into tissues, it also facilitates the fungal endocytosis through the same mechanism. Moreover, upregulation of GRP78 by viral spike protein is not only limited to the internalization of Mucorales into the endothelium, but also enhances its pathogenicity and virulence [49, 54].

Notably, studies have shown that the antibodies against GRP78 suppress the fungal invasion and drastically reduce the endothelial injury in a susceptible host [20]. In addition to that, GRP78 antibodies have been shown to protect diabetic ketoacidotic mice from fungal disease indicating a major role of GRP78 in the pathogenesis [54]. Therefore, it can be substantiated that SARS-CoV-2 infection independently increases the risk factor for mucormycosis by upregulating the GRP78. As per the literature, GRP78 is known to be utilized for internalization by other viruses also such as Ebola and Influenza virus and co-incidentally, and association of mucormycosis with these viruses has been documented in a few case reports further increasing the credibility of these findings [55–57].

Since GRP78 is stress-related protein, interaction with elevated concentrations of blood glucose and ketone bodies on the endothelium also enhances its expression [20, 48]. Furthermore, SARS-CoV-2 infection results in an increase in circulating heme, iron and ferritin as a result of its pathogenesis as mentioned earlier. Consequently, this may result in free radical injury and contributes to endothelial wall remodeling resulting in diffuse endothelitis [7]. This diffuse endothelitis is an independent risk factor, and it upregulates the GRP78 and also catalyzes the adhesion of *R. Oryzae* to the endothelium via GRP-78 [48].

Studies reveal that elevated GRP78 expression and subsequent CotH3 interaction enhance the virulence of the fungus, thereby increasing its ability to invade and damage endothelium in a receptor-dependent manner [49, 54]. It has been shown that the *Rhizopus Oryzae* with attenuated expression of CotH3 demonstrated significant reduction in tissue invasion, endothelial damage and virulence in DKA

murine model of mucormycosis [54]. More recently, other studies have shown the GRP78-dependent endothelial invasion of the otherwise noninvasive saccharomyces cerevisiae by expressing CotH genes which further supports the role of GRP78/CotH interaction in the pathogenesis of *R. Oryzae* [49]. Interestingly, the CotH3 is exclusively present in all Mucorales but absent from other opportunistic fungi like Candida and Aspergillosis, a likely explanation for increased susceptibility of these population to mucormycosis.

Finally, in accordance with the GRP78 expression by free iron and glucose levels, it was found that the mice with induced DKA were found to express GRP78 mRNA, 2–5 times higher than the normal mice particularly in organs like sinus lining and brain [54]. In this finding, the overexpression of GRP78 in the sinus lining can be principally translated as, firstly to the present-day surge in the cases of rhino-cerebral disease in SARS-CoV-2 and secondly, the predisposition of diabetic ketoacidosis to the rhino-cerebral disease. Indeed, these findings are remarkable and indicative of the therapeutic potential of GRP78-blocking strategies in treating or preventing mucormycosis in SARS-CoV-2 pandemic.

Taking into account that the endothelitis and oxidative stress in the vascular endothelium are the independent risk factors, statins have a significant role in the primary prevention of mucormycosis. Surprisingly, quite a number of studies have documented the benefits of statins in inhibiting the growth of Zygomyces [58]. The pleiotropic effects of statins make this class of drugs highly effective in diseases where innate immunity and endothelial damage play a key role.

Bellanger et al. [58] demonstrated that the exposure to statins at concentration below the MICs not only induced apoptosis by DNA fragmentation [antifungal activity] but also markedly reduced the ability of *R. oryzae* to adhere, invade and damage the endothelial cells. Although future studies are needed for clinical translation of the results, using statins as prophylaxis, adjunctive therapy or in combination with conventional antifungal agents should be considered favorably in the management of mucormycosis. The lower incidence of mucormycosis in developed countries is presumably because of the liberal of the use of statins in metabolic syndromes as speculated by various authors [2, 59].

Accurately summarizing the above, angioinvasion is a consequence of the unique interaction between the fungus CotH3 ligand and endothelium GRP78. Strikingly, the factors which upregulate the expression of GPR78 and CotH ligand culminating in fungal invasion and growth include DKA, high serum glucose, endothelitis, systemic inflammation, ferritin and elevated serum Iron. Fundamentally, all the above factors can either be directly or indirectly linked to of SARS-CoV-2 infection or its pathogenesis. Fitting with this concept and the uniformity of the reports of mucormycosis in patients with COVID-19 implicate a robust link in the pathogenesis of mucormycosis and COVID-19 infection.

Mucormycosis as a Nosocomial Infection

The occurrence of mucormycosis as a nosocomial infection is rare. Recently, the use of water from overhead water-tanks in humidifier has been quoted as a cause of Mucormycosis; however, this theory may not be entirely true. It is debatable, why different genera of Mucor fungi are isolated from different patients treated in the same hospital at a particular time (a single pathogen is usually the cause for nosocomial outbreak). Similarly, the humidifier theory fails to explain, why the spores reaching the lungs via water/air are not causing any infection in the compromised lungs, but are resulting in only the rhino-cerebral variant of mucormycosis. So far only two cases of pulmonary mucormycosis have been reported in conjunction with the pandemic [60]. Hence, the occurrence of isolated rhino-cerebral variant without any lung infection completely refutes this observation. Additionally, as per the literature the incubation period of rhino-cerebral mucormycosis is 2–5 days [61]. Conforming to this, all infections should commence within 5 days of admission to the hospital. This is not true as majority of mucormycosis associated with COVID-19 is observed weeks after the apparent recovery of the patient.

It has also been postulated that oxygen cylinders (industrial oxygen) or oxygen concentrators could be contaminated with spores. Since mucormycosis is an epidemic within a pandemic, the whole Indian subcontinent has been affected. Hence, it is not logical to believe that all the oxygen supplies in the country are distributed from the same contaminated plant or that all the oxygen-generating plants in our nation are infested with Mucorales. Similarly, it does not explain why the oxygen contaminated with fungal spores is causing exclusively rhino-cerebral variant and not pulmonary.

References

1. Roden MM, Zaoutis TE, Buchanan WL, Knudsen TA, Sarkisova TA, Schaufele RL et al (2005) Epidemiology and outcome of zygomycosis: a review of 929 reported cases. Clin Infect Dis 41:634–653
2. Petrikkos G, Skiada A, Lortholary O, Roilides E, Walsh TJ, Kontoyiannis DP (2012) Epidemiology and clinical manifestations of mucormycosis. Clin Infect Dis 54(Suppl 1):S23–S34
3. Chakrabarti A, Singh R (2014) Mucormycosis in India: unique features. Mycoses 57(Suppl. 3):85–90

4. Petrikkos G, Skiada A, Lortholary O et al (2012) Epidemiology and clinical manifestations of mucormycosis. Clin Infect Dis 54:S23–S34
5. Edeas M, Saleh J, Peyssonnaux C (2020) Iron: innocent bystander or vicious culprit in COVID-19 pathogenesis? Int J Infect Dis 97:303–305
6. Sharma S, Grover M, Bhargava S, Samdani S, Kataria T (2021) Post coronavirus disease mucormycosis: a deadly addition to the pandemic spectrum. J Laryngol Otol, 1–6
7. Cavezzi A, et al (2020) COVID-19: hemoglobin, iron, and hypoxia beyond inflammation: a narrative review. Clin Pract
8. Spellberg B, Edwards J Jr, Ibrahim A (2005) Novel perspectives on mucormycosis: pathophysiology, presentation, and management. Clin Microbiol Rev 18:556–569
9. Moorthy A, Gaikwad R, Krishna S, Hegde R, Tripathi KK, Kale PG, Rao PS, Haldipur D, Bonanthaya K (2021) SARS-CoV-2, uncontrolled diabetes and corticosteroids-an unholy trinity in invasive fungal infections of the maxillofacial region? A retrospective multi-centric analysis. J Maxillofac Oral Surg 6:1–8
10. John TM, Jacob CN, Kontoyiannis DP (2021) When uncontrolled diabetes mellitus and severe COVID-19 converge: the perfect storm for mucormycosis. J Fungi 7:298
11. Guan W-J, Ni Z-Y, Hu Y, Liang W-h, Ou C-Q, He J-X et al (2020) Clinical characteristics of 2019 novel coronavirus infection in China. medRxiv
12. Li G, et al (2020) Coronavirus infections and immune responses. J Med Virol
13. Antinori S, Nebuloni M, Magni C, Fasan M, Adorni F, Viola A, Corbellino M, Galli M, Vago G, Parravicini C et al (2009) Trends in the postmortem diagnosis of opportunistic invasive fungal infections in patients with AIDS: a retrospective study of 1630 autopsies performed between 1984 and 2002. Am J Clin Pathol 132:221–227
14. Bouchara JP, Oumeziane NA, Lissitzky JC, Larcher G, Tronchin G, Chabasse D (1996) Attachment of spores of the human pathogenic fungus Rhizopus oryzae to extracellular matrix components. Eur J Cell Biol 70(1):76–83
15. Koparal M, Kurt E, Altuntas EE, Dogan F (2021) Assessment of mucociliary clearance as an indicator of nasal function in patients with COVID-19: a cross-sectional study. Eur Arch Otorhinolaryngol 278(6):1863–1868
16. Çeçen A, Bayraktar C, Özgür A, Akgül G, Günal Ö (2021) Evaluation of nasal mucociliary clearance time in COVID-19 patients. J Craniofacial Surg: April 29, 2021—volume Publish Ahead of Print - Issue
17. Chang D, Lin M, Wei L, Xie L, Zhu G, Dela Cruz CS, Sharma L (2020) Epidemiologic and clinical characteristics of novel coronavirus infections involving 13 patients outside Wuhan, China. JAMA
18. Ibrahim AS, Voelz K (2017) Themucormycete–host interface. Curr Opin Microbiol 40:40–45
19. Lortholary O, Renaudat C, Sitbon K, Madec Y, Denoeud-Ndam L, Wolff M, Fontanet A, Bretagne S, Dromer F; French Mycosis Study Group (2014) Worrisome trends in incidence and mortality of candidemia in intensive care units [Paris area, 2002–2010]. Intensive Care Med 40(9): 1303–1312
20. Gebremariam T, Lin L, Liu M et al (2016) Bicarbonate correction of ketoacidosis alters host-pathogen interactions and alleviates mucormycosis. J ClinInvest 126:2280–2294
21. Spellberg B, Andes D, Perez M, Anglim A, Bonilla H, Mathisen GE et al (2009) Safety and outcomes of openlabel deferasirox iron chelation therapy for mucormycosis. Antimicrob Agents Chemother 53(7):3122–3125
22. Oriot P, Hermans MP (2020) Euglycemic diabetic ketoacidosis in a patient with type 1 diabetes and SARS-CoV-2 pneumonia: case report and review of the literature. Acta Clin Belg
23. Smith SM, Boppana A, Traupman JA, Unson E, Maddock DA, Chao K, Dobesh DP, Brufsky A, Connor RI (2021) Impaired glucose metabolism in patients with diabetes, prediabetes, and obesity is associated with severe COVID-19. J Med Virol 93(1):409–415
24. Wu L, Girgis CM, Cheung NW (2020) COVID-19 and diabetes: insulin requirements parallel illness severity in critically unwell patients. Clin Endocrinol 93:390–393
25. Gianchandani R et al (2020) Managing hyperglycemia in the COVID-19 inflammatory storm. Diabetes 69:2048–2053
26. Pal R, Banerjee M, Yadav U, Bhattacharjee S (2020) Clinical profile and outcomes in COVID-19 patients with diabetic ketoacidosis: a systematic review of literature. Diabetes Metab Syndr 14:1563–1569
27. Potier L et al (2020) COVID-19 symptoms masking inaugural ketoacidosis of type 1 diabetes. Diabetes Metab
28. Marchand L, Pecquet M, Luyton C (2020) Type 1 diabetes onset triggered by COVID-19. Acta Diabetol 57:1265–1266
29. Yang JK, Lin SS, Ji XJ, Guo LM (2010) Binding of SARS coronavirus to its receptor damages islets and causes acute diabetes. Acta Diabetol 47(3):193–199
30. Kontoyiannis DP, Lewis RE (2011) How I treat mucormycosis. Blood 118:1216–1224
31. Min KH, Rhee CK, Jung JY, Suh MW (2012) Characteristics of adverse effects when using high dose short term steroid regimen. Korean J Audiol 16(2):65–70
32. Howard DH (1999) Acquisition, transport, and storage of iron by pathogenic fungi. Clin Microbiol Rev 12:394–404
33. Artis WM, Fountain JA, Delcher HK, Jones HE (1982) A mechanism of susceptibility to mucormycosis in diabetic ketoacidosis: transferrin and iron availability. Diabetes 31:1109–1114
34. Majeed A, Shajar MA (2020) Is hemoglobin the missing link in the pathogenesis of COVID-19? Anaesth Pain Intensive Care 24:9–12
35. Chen C, et al (2020) Advances in the research of cytokine storm mechanism induced by Corona Virus Disease 2019 and the corresponding immunotherapies, Chin. J Burns
36. Colafrancesco et al (2020) COVID-19 gone bad: a new character in the spectrum of the hyperferritinemic syndrome? Autoimmunity reviews
37. Merad M, Martin J (2020) Pathological inflammation in patients with COVID-19: a key role for monocytes and macrophages. Nat Rev Immunol. /https://doi.org/10.1038/S41577-020-0331-4
38. Perricone C, Bartoloni E, Bursi R, Cafaro G, Guidelli GM, Shoenfeld Y, Gerli R (2020) COVID-19 as part of the hyperferritinemic syndromes: the role of iron depletion therapy. Immunol Res 68:213–224
39. Pretorius E, Kell D (2014) Diagnostic morphology: biophysical indicators for iron-driven inflammatory diseases. Integr Biol [Camb]
40. Goldberg M et al (2020) Cerebrovascular disease in COVID-19. Am J Neuroradiol
41. Varga Z, Flammer AJ, Steiger P et al (2020) Endothelial cell infection and endothelitis in COVID-19. Lancet 395:1417–1418
42. Ganz T, Nemeth E (2012) Hepcidin and iron homeostasis. Biochim Biophys Acta 1823:1434–1443
43. Ehsani S (2003) Distant sequence similarity between hepcidin and the novel coronavirus spike glycoprotein: a potential hint at the possibility of local iron dysregulation in COVID-19. arXiv: 2003
44. Hirschhorn T, Stockwell BR (2019) The development of the concept of ferroptosis. Free Radic Biol Med 133:130–143
45. Shirazi F, Kontoyiannis DP, Ibrahim AS (2015) Iron starvation induces apoptosis in Rhizopus oryzae in vitro. Virulence 6:121–126

46. Spellberg B, Ibrahim AS, Chin-Hong PV, Kontoyiannis DP, Morris MI, Perfect JR et al (2012) The deferasirox- AmBisome therapy for mucormycosis [DEFEAT Mucor] study: a randomized, double-blinded, placebo-controlled trial. J Antimicrob Chemother 67(3):715–22
47. Ibrahim AS, Gebermariam T, Fu Y, Lin L, Husseiny MI, French SW et al (2007) The iron chelator deferasirox protects mice from mucormycosis through iron starvation. J Clin Invest 117(9):2649
48. Baldin C, Ibrahim AS (2017) Molecular mechanisms of mucormycosis—the bitter and the sweet. Sheppard DC, ed. PLOS Pathog 13: e1006408
49. Gebremariam T, Liu M, Luo G, Bruno V, Phan QT, Waring AJ, Edwards JE, Filler SG, Yeaman MR, Ibrahim AS (2014) CotH3 mediates fungal invasion of host cells during mucormycosis. J Clin Investig 124:237–250
50. Ibrahim IM, Abdelmalek DH, Elfiky AA (2019) GRP78: a cell's response to stress. Life Sci 226:156–163
51. Ibrahim IM, Abdelmalek DH, Elshahat ME, Elfiky A (2020) COVID-19 spike-host cell receptor GRP78 binding site prediction. J Infect 8:554–562
52. Fu J, Wei C, He J, Zhang L, Zhou J, Balaji KS, Shen S, Peng J, Sharma A, Fu J (2021) Evaluation and characterization of HSPA5 [GRP78] expression profiles in normal individuals and cancer patients with COVID-19. Int J Biol Sci 17(3):897–910
53. Belyamani L, Ibrahimi A (2020) Targeting the GRP78-dependant SARS-CoV-2 cell entry by peptides and small molecules. Bioinform Biol Insights 21(14):11
54. Liu M, Spellberg B, Phan QT et al (2010) The endothelial cell receptor GRP78 is required for mucormycosis pathogenesis in diabetic mice. J Clin Invest 120:1914–1924
55. Kalongi Y, Mwanza K, Tshisuaka M, Lusiama N, Ntando E, Kanzake L, Rollin P (1999) Isolated case of ebola hemorrhagic fever with mucormycosis complications, Kinshasa, Democratic Republic of the Congo. J Infect Dis 179:, S15-S17
56. Ahmadikia K, Hashemi SJ, Khodavaisy S et al (2021) The double-edged sword of systemic corticosteroid therapy in viral pneumonia: a case report and comparative review of influenza-associated mucormycosis versus COVID-19 associated mucormycosis. Mycoses 00:1–11
57. Sabirli R, Koseler A, Goren T, Turkcuer I, Kurt O (2021) High GRP78 levels in Covid-19 infection: a case-control study. Life Sci 265: 118781
58. Bellanger AP, Tatara AM, Shirazi F et al (2016) Statin concentrations below the minimum inhibitory concentration attenuate the virulence of Rhizopus oryzae. J Infect Dis 214:114–121
59. Reed C, Bryant R, Ibrahim AS et al (2008) Combination polyene-caspofungin treatment of rhino-orbital-cerebral mucormycosis. Clin Infect Dis 47:364–371
60. Garg D, Muthu V, Sehgal IS et al (2021) Coronavirus disease (Covid-19) associated mucormycosis (CAM): case report and systematic review of literature. Mycopathologia 186:289–298
61. Smith RM, Lee J, Mody RK (2015) Determining the incubation time of mucormycosis: a systematic review. Open Forum Infectious Diseases, Volume 2, Issue suppl_1, 461